

## MAPKs activation and mitochondrial depletion are associated with chemotherapy-related cachexia

Rafael Barreto<sup>1</sup>, David L. Waning<sup>2,3,5</sup>, Hongyu Gao<sup>4</sup>, Yunlong Liu<sup>4</sup>, Teresa A. Zimmers<sup>1,2,5</sup>, Andrea Bonetto<sup>1,2,5</sup>

<sup>1</sup>Department of Surgery, <sup>2</sup>Simon Cancer Center, <sup>3</sup>Department of Medicine, <sup>4</sup>Department of Medical and Molecular Genetics, Center for Computational Biology and Bioinformatics, Indiana University School of Medicine, Indianapolis, IN 46202, USA; <sup>5</sup>IUPUI Center for Cachexia Research, Innovation and Therapy, Indianapolis, IN 46202, USA

**Background.** Cachexia, defined by increased fatigue and loss of muscle function, results from muscle and fat depletion and affects the majority of cancer patients with no effective treatments. Previous studies suggest that chemotherapy itself may contribute to cachexia, although the mechanisms responsible for these derangements are not clear. The purpose of this study was to investigate the mechanism(s) associated with chemotherapy-related effects on body composition and muscle function.

**Methods.** Chemotherapy regimens routinely used for the therapy of solid tumors were tested in normal CD2F1 mice, followed by assessment of body composition and muscle strength. Mitochondrial activity in muscle sections was evaluated, and TEM imaging in EDL muscle was performed. To determine whether chemotherapy modulates signaling pathways associated with the regulation of muscle mass, Western blotting, qRT-PCR and RNA-Sequencing were utilized.

**Results.** Administration of Folfox (5-FU, leucovorin, oxaliplatin), Folfiri (5-FU, leucovorin, irinotecan) or Gemcitabine/Paclitaxel for up to 5 weeks to normal mice caused marked decreases in adipose tissue and skeletal muscle content, coherent with reduced muscle strength. Notably, ERK1/2/MAPK and p38/MAPK signaling pathways, as well as myostatin expression were significantly up-regulated. TEM analysis unveiled a marked depletion in muscle mitochondrial content and alterations of the sarcomeric structure consistent with loss of muscle structural proteins in the mice receiving chemotherapy. Moreover, the RNA-Sequencing analysis identified several markers associated with mitochondrial homeostasis, lipid metabolism and acute phase response that were significantly affected by Folfiri administration.

**Conclusions.** Our findings suggest that chemotherapy promotes the activation of MAPK- and myostatin-dependent muscle atrophy and causes mitochondrial depletion and alterations of the sarcomeric units, likely playing a causative role in the occurrence of muscle loss and weakness. Future investigations will clarify whether pharmacologically increasing muscle mass or inhibiting MAPK activation reduces chemotherapy-related cachexia, thereby providing potential pharmacological targets to improve efficacy and tolerance of anticancer drugs.